New information on the genus *Pseudoganisa* SCHULTZE, 1910 (Lepidoptera: Eupterotidae): mtDNA barcode indicates two cryptic species on the Philippines

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Abstract: The formerly monotypic genus Pseudoganisa SCHULTZE, 1910, an endemic to the Philippine Archipelago, was found during serial mtDNA-COI-barcode scanning to consist of at least 2 different cryptic species (over 6% difference in sequence data): P. currani Schultze, 1910, from Mindanao, and P. gonioptera (West, 1932), stat. rev., from the northern islands. This result is in contrast to an earlier publication (Nässig & Treadaway 2009) based on morphology alone. The northern P. gonioptera appears to be rather heterogenous, with slight differences between the islands studied. The distribution is mapped, the newly grouped taxa and their morphology shortly described and differentiated, with illustrations of some of their genitalia, and the barcode data are shown. The new results were primarily obtained with mtDNA (COI) barcode data and statistic support (ME-GA5); a later support by morphological data was hardly possible due to extreme variability (individual and also, at a smaller degree, between islands) and only based on small and inconspicuous charcters.

Neue Informationen über die Gattung *Pseudoganisa* SCHULTZE, 1910 (Lepidoptera: Eupterotidae): Der mt-DNA-Barcode zeigt zwei kryptische Arten an

Zusammenfassung: Die zuvor als monotypisch betrachtete Gattung Pseudoganisa Schultze, 1910, ein Endemismus der Philippinischen Inseln, wurde im Rahmen einer Reihenuntersuchung des mtDNA-COI-"Barcodes" als aus mindestens zwei deutlich unterschiedenen (über 6% Unterschied in den Basenpaarsequenzdaten) kryptischen Arten bestehend erkannt: P. currani Schultze, 1910 von Mindanao und P. gonioptera (West, 1932), stat. rev., von den nördlicheren Inseln. Dieses Ergebnis steht in Kontrast zu einer früheren, ausschließlich auf morphologischen Daten basierenden Publikation (Nässig & Treadaway 2009). Die nördlichen Populationen von P. gonioptera sind offenbar sehr heterogen und zeigen hohe individuelle und geografische Variabilität, die eine morphologische Differenzierung von der südlichen Art kaum möglich machen. Die Verbreitung der beiden Arten (soweit bisher geklärt) wird auf einer Karte dargestellt, die beiden neu unterteilten Arten werden kurz beschrieben, einige männliche Genitalien und berechneten Barcode-Bäumen abgebildet. Die neuen Ergebnisse beruhen in erster Linie auf Analysen des mtDNA-(COI-)"Barcodes" und statischer Software (MEGA5).

Introduction

Not long ago, Nässig & Treadaway (2009) provided a review over the endemic Philippine genus *Pseudoganisa* Schultze, 1910, being a member of the informal "*Ganisa*-group" of the family Eupterotidae. This informal group name was proposed by Oberprieler et al. (2003)

and Nässig & Oberprieler (2007, 2008) for a number of genera of still unclear phylogenetic placement within the family. The higher systematics (at the family, subfamily and tribal levels) and phylogeny of the family Eupterotidae are still not reliably resolved and are at present under research (see Oberprieler et al. 2003, Nässig & Oberprieler 2007, 2008, Zwick 2008, Zwick et al. 2011).

In their publication, Nässig & Treadaway (2009) came to the morphology-based conclusion that all populations from all islands studied belong to a single species, *Pseudoganisa currani* Schultze, 1910 (described from "Mindanao, District of Zamboanga"), with *Apha gonioptera* West, 1932 (described from "Luzon, subprov. Benguet, Palali") being no more than a synonym of the first taxon. This interpretation was our opinion at that time notwithstanding the observation that there were many differences in \eth genitalia, but combined with what was then seen as a large individual variability. The same synonymy was already (although then only somewhat preliminarily and tentatively) proposed by Nässig (1995).

Shortly after that publication of 2009, we began a general scanning of mt-DNA COI-5p barcodes (see BARCODE OF Life 2013) of Asian Eupterotidae, initially starting with material mainly from the private collection of one of the present authors (S.L., with support by S.N.). The barcode sequences of the Eupterotidae in the collections of the Senckenberg-Museum (SMFL, including coll. W. A. Näs-SIG = CWAN) and in the private collection of C. G. TREAD-AWAY (CCGT, assigned to SMFL) have, with a few exceptions, not yet been included in that study, because the conditions for obtaining DNA-barcode results in Canada were changed abruptly and without prior information in early 2012 prohibiting further wide "screening" studies across three bombycoid families (Saturniidae, Brahmaeidae and Eupterotidae) without funding; so we have not yet achieved the screening of the mt-DNA of all insular populations available to us.

The initial results of these barcode studies were slightly surprising with respect to *Pseudoganisa*, showing unexpected large differences in the nucleotid sequences (over 6%) between specimens from Mindanao and all other islands of the Philippines studied so far. Consequently, the new data available to us now shows a more differentiated picture than before which we publish here, as we do not see a realistic chance to complete our COI barcode

¹ Studies in Eupterotidae (Eupterotiden-Studien), no. 13. (No. 12 see: Nässig, W. A., Holloway, J. D., & Beeke, M. (2011): Description of a new species of *Eupterote* (Eupterote) from Sulawesi (Indonesia) (Lepidoptera: Eupterotidae). — Nachrichten des Entomologischen Vereins Apollo, Frankfurt am Main, N.F. 31 (4): 197–200.)

studies at present or within a short time. Especially island records based on very small series or singletons have not been barcoded so far, as have similar groups from a few of the larger islands.

Our mt-DNA studies also indicated that our expectation from earlier morphological studies that *Pseudoganisa* appears to be the sister-group to the mainly continental genus *Apha* Walker, 1855 can not only be supported based on genitalia and external morphology, but also using COI barcode results; therefore, we used two specimens of *Apha* of two different species as outgroup for *Pseudoganisa* here in our DNA study.

Methods

General abbreviations and conventions see in Nässig & Treadaway (2009). DNA barcodes (658 base pairs = bp of the COI mitochondrial gene) were generated following the routine procedure at the Canadian Centre for DNA Barcoding (CCDB 2013) as detailed, e.g., in Vaglia et al. (2008).

DNA was extracted from the legs of dried specimens in the collections of two of the authors (S.L. and S.N.). A total of 8 specimens of *Pseudoganisa* (plus 2 specimens of *Apha* as outgroup) were processed (see Tab. 1). The sequence data are made public via the BOLD website and GenBank. The statistically analysed number of base pairs was only 631, because not all specimens resulted in a complete analysis of the full DNA length of 658 bp.

Results

Barcode analysis

1. The Palawan specimen

The zoogeographically exceptional record of a single Q in CSLL labelled as collected in xi. 2004 by a Philippine insect trader on Palawan (see Nässig & Treadaway

2009: fig. 3) was supported by COI barcode analysis. The specimen definitively belongs to Pseudoganisa gonioptera, as the first known (and so far only) record of the genus from the lands on the Sunda shelf. The barcode analysis took nearly a year between submission of the leg in late 2011 in Canada, followed by a first unsuccessful analysis (no sequence data resulting), and summer 2012, when after failure tracking and repeating the analysis with shorter primers the sequence data was eventually showing up on the BOLD database. For many years it was assumed that the distribution area of Pseudoganisa excludes the zoogeographical region of Palawan (see Nässig & Oberprieler 2008: 63), which politically belongs to the Philippines but geologically is part of Sundaland (Vane-Wright 1990). Therefore, the DNA-based confirmation of the probably autochthonous origin for Palawan of this singleton is a valuable result.

2. Intrageneric results

The sequence data were analysed with the software MEGA5 (Tamura et al. 2011), and the resulting Maximum Likelihood (ML) (with, in addition, a linearized variant of it in Fig. 1b, to show more clearly the percentage of differences in sequences) and, for comparison, Minimum Evolution (ME) consensus trees are illustrated in Figs. 1a–1c; for further details of the parameter used in calculating the statistics see legend. Further methods offered by MEGA5 software resulted in quite similar topographies.

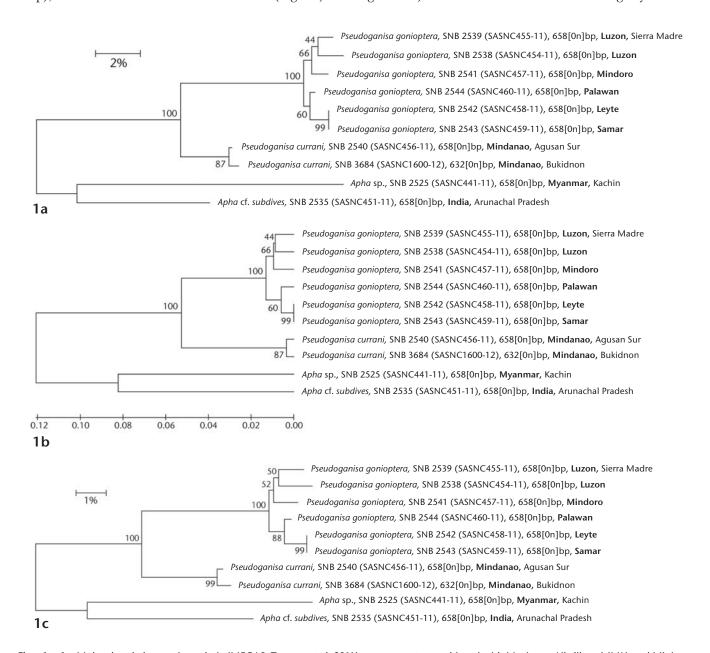
The results show a clear separation (with over 6% of the sequence data) between the specimens from Mindanao and all other specimens. For these two, different names are available (see below, chapter Systematics).

There is some more "topological structure" in the northern species, with the specimens from Palawan, Samar and Leyte islands being somehow separate from the

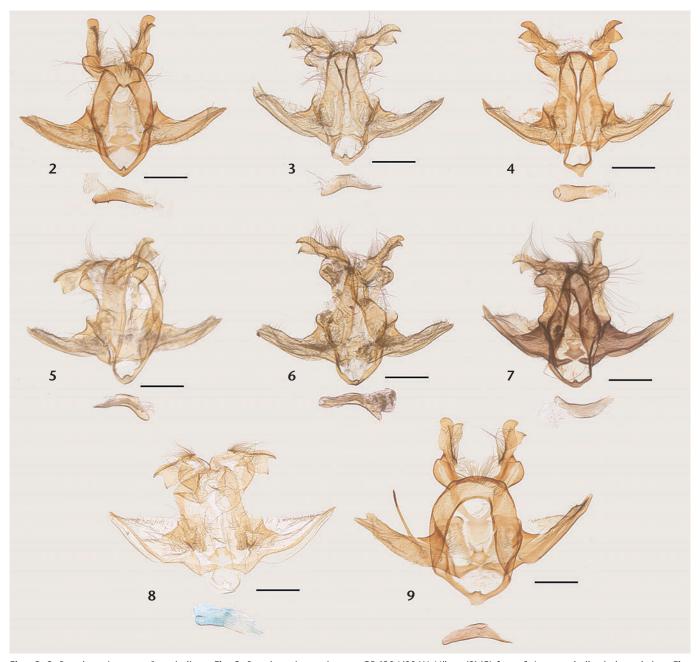
Tab. 1: Specimen and barcode data, locality of collecting and depository for the specimens used here in the publication. — For general abbreviations see Nässig & Treadaway (2009). Collection acronyms: CSLL =coll. S. LÖFFLER; CSNB = coll. S. NAUMANN. Additional abbreviations: BC = barcode no.; GBAC = GenBank Access Code; SL = COI-5P sequence length in base pairs; — = GBAC not yet available.

Taxon	ВС	BOLD Process ID	GBAC	Sex	SL	Locality	Coll.	
Pseudoganisa currani	SNB 2540	SASNC456-11	JX681812	ð	658 [0n]	Philippines, Mindanao, Agusan Sur, 10 km SE Trento Sta. Maria, 185 m, 27. iv. 2008, leg. JHL (8°1.615' N, 126°12.322' E)		
Pseudoganisa currani	SNB 3684	SASNC1600-12	JX681809	9	632 [0n]	Philippines, Mindanao, Bukidnon, NW Maramag, 28. ix. 2011, leg. SN & JHL		
Pseudoganisa gonioptera	SNB 2538	SASNC454-11	JX681811	ð	658 [0n]	Philippines, Luzon, 5 km S Adams, 350 m, 6. iv. 2008, leg. JHL (18°31.338′ N, 120°55.690′ E)		
Pseudoganisa gonioptera	SNB 2539	SASNC455-11	JX681810	3	658 [0n]	Philippines, Luzon, Sierra Madre, Aurora, 585 m, 5. ix. 2007, leg. JHL (16°32.886′ N, 122°14.134′ E)	CSLL	
Pseudoganisa gonioptera	SNB 2541	SASNC457-11	JX681808	3	658 [0n]	Philippines, Mindoro, Mt. Halcon, 1000 m, 2. viii. 2000, le N. Mohagan		
Pseudoganisa gonioptera	SNB 2543	SASNC459-11	JX681806	3	658 [0n]	Philippines, Samar, Mt. Capoto-an, 600 m, 19. i. 2005, leg. R. Cabale		
Pseudoganisa gonioptera	SNB 2542	SASNC458-11	JX681807	ð	658 [0n]	Philippines, Leyte, Mt. Balocaue, 1. v. 2001, leg. loc. coll.	CSLL	
Pseudoganisa gonioptera	SNB 2544	SASNC460-11	JX681813	\$	658 [0n]	Philippines, Palawan, Mt. Mantalingajan, Brooke's Point, 15. xi. 2004, leg. I. Fierro		
				Out	group			
Apha cf. subdives	SNB 2536	SASNC451-11	_	ð	658 [0n]	India, Arunachal Pradesh, Distr. Along, nr. Rapum, 2000 m, 6. vii. 2010, leg. G. Bretschneider		
Apha sp.	SNB 2525	SASNC441-11	JX681805	₫	658 [0n]	Myanmar, Kachin, Chudu Razi Hills, 30 miles E Kawnglangphu, 12. vii. 2007, leg. loc. coll.	CSLL	

rest, and also nearly every insular population studied so far being slightly separate for itself. However, there are so few DNA sequence data available (the COI-barcode of so many insular populations is still unknown, see map), and as shown in the linearized tree (Fig. 1b) the sequence difference percentages among the northern insular populations are much smaller than the difference between these and the Mindanao population. Further, the results of the morphological studies (especially \eth genitalia) are still rather indifferent and slightly obscure.



Figs. 1a-1c: Molecular phylogenetic analysis (MEGA5, TAMURA et al. 2011), consensus trees achieved with Maximum Likelihood (ML) and Minimum Evolution (ME) methods. The analyses involved 10 nucleotide sequences (= specimens), 8 of the genus Pseudoganisa, 2 of Apha as outgroup. There were a total of 631 positions in the final dataset, all positions containing gaps and missing data were eliminated. - Fig. 1a: The evolutionary history was inferred by using the Maximum Likelihood method based on the data specific model (NEI & KUMAR 2000). The bootstrap consensus tree inferred from 1000 replicates (Felsenstein 1985) is taken to represent the evolutionary history of the taxa analyzed. The percentage of replicate trees in which the associated taxa clustered together in the bootstrap test are shown next to the branches. The tree with the highest log likelihood (-1672.3830) is shown. An initial tree for the heuristic search was obtained automatically using the maximum parsimony method, because the number of common sites was < 100 or less than one fourth of the total number of sites. A discrete Gamma distribution was used to model evolutionary rate differences among sites (5 categories [+ G, parameter = 8.1177]). The rate variation model allowed for some sites to be evolutionarily invariable ([+ I], 67.26% sites). The tree is drawn to scale, with branch lengths measured in the number of substitutions per site. Codon positions included were 1st + 2nd + 3rd + Noncoding. - Fig. 1b: The same ML-consensus tree as before, but linearized, so that the percentage of difference in the sequences becomes more clearly visible. — Fig. 1c: The evolutionary history was inferred using the Minimum Evolution method (RZHETSKY & NEI 1992). The percentage of replicate trees in which the associated taxa clustered together in the bootstrap test (1000 replicates) are shown next to the branches (FELSENSTEIN 1985). The tree is drawn to scale, with branch lengths in the same units as those of the evolutionary distances used to infer the phylogenetic tree. The evolutionary distances were computed using the Maximum Composite Likelihood method (TAMURA et al. 2004) and are in the units of the number of base substitutions per site. The rate variation among sites was modeled with a gamma distribution (shape parameter = 3). The differences in the composition bias among sequences were considered in evolutionary comparisons (TAMURA & KUMAR 2002). The ME tree was searched using the Close-Neighbor-Interchange (CNI) algorithm (NEI & KUMAR 2000) at a search level of 0. The Neighbor-joining algorithm (SAITOU & NEI 1987) was used to generate the initial tree. - Similar topographies of trees were also achieved applying other methods offered by MEGA5: UPGMA and Neighbor Joining.



Figs. 2–9: Pseudoganisa spp., ♂ genitalia. — Fig. 2: Pseudoganisa gonioptera, GP 1304/00 W. Nässig/SMFL [wng.]; Luzon; phallus in lateral view. Fig. 3: Ps. gonioptera, GP 2035/08 wng.; Mindoro; phallus lateral. Fig. 4: Ps. ?gonioptera [population from this island not barcoded], GP 1305/00 wng.; Negros; phallus ventral. Fig. 5: Ps. ?gonioptera [not barcoded], GP 2030/08 wng.; Cebu; phallus lateral. Fig. 6: Ps. gonioptera, GP 2031/08 wng.; Leyte; phallus lateral. Fig. 7: Ps. ?currani [not barcoded], GP 2028/08 wng.; Jolo; phallus lateral. Fig. 8: Ps. sp.? [not barcoded], GP 11.581 Witt; "Sulawesi" [sic, erroneous locality, the genus Pseudoganisa is not known from any Indonesian island!]; phallus lateral, specimen in coll. Witt. Fig. 9: Ps. currani, GP no. 1306/00 W. Nässig/SMFL; Mindanao; phallus lateral. — Scale bars = 1 mm, pictures approximately to the same size. Genitalia scans through support by W. Eckweiler, Frankfurt am Main.

So we decided not to split up this northern population any further for the time being (in spite of some high bootstrap values in the statistical analysis). There is no reliable morphological support so far, and much more research is required.

Morphological differentiation

Both species, *Pseudoganisa currani* as well as *P. gonioptera*, are externally very variable species, see the colour plates in Nässig & Treadaway (2009). There is, in addition, some moderate sexual dimorphism (especially in wingshape), but colour and pattern (and their variability) are similar (and vary similarly) in both sexes.

There is, however, no clear morphological separation between the species, based on external morphology only; variability ranges, sizes and other parameters overlap to differing degrees. Further, the high variability of the \eth genitalia (see Figs. 2-9) does not support a reliable and safe separation of the two species. At present, it is not yet clear whether the insular variability is dominating the individual or vice versa.

So, at present we cannot safely provide reliable characters for separating the species based on singletons and external or genitalia morphology except by locality data and COI barcode analysis. Only in longer series slight differences between species and islands can sometimes be

detected, but remain inconspicuous. Further studies of genitalia morphology, combined with COI barcode, on long series of specimens remain necessary.

The valves of the 3 genitalia of both species are unique in the family Eupterotidae (see Nässig & Treadaway 2009): the apical part of the costal lobe is rolled ventrally like an envelope or sheath around the sclerotised, needle-like apical part of the sacculus; this sheath is open near base but almost closed near apex (but open at the most distal tip), sometimes even appearing fused to a tube. Also, there are well-developed tufts of pheromone hairs laterally in the intersegmental fold between the 3 genitalia and the 8th abdominal segments, inserting ventrally and stretching more or less along the valves on their outside. However, these are quite loose and usually get lost during the dissection process.

Distribution, ecology and other observations

Variability and distribution. The smallest specimens are known from Luzon (15 $\eth \eth$, 1 \circlearrowleft) in the north and Jolo (n=1 \eth) in the south; specimens from the West Visayan and Mindanao regions usually are the largest (Nässig & Treadaway 2009: tab. 1). Therefore, size is also no sufficient character to distinguish both species. The variability in colour and pattern appears to be somehow correlated with collecting dates (i.e., season) rather than locality (i.e., different species or populations and geography). There is obviously no pattern or colour combination particular to any one area and species.

Phenology. All specimens were collected at light, and obviously both sexes are nocturnal. For the collecting months see Tab. 2. Both species appear to be multivoltine, having been collected in nearly all months of the year, but as the regional dry and wet seasons are quite variable and different within the Philippines (even within the larger islands, depending on mountain ranges and prevailing seasonal winds), further collecting data may possibly disclose some regional seasonality of the two

species within the archipelago. Such studies may best be conducted by researchers living somewhere in the Philippine Archipelago.

Both species of *Pseudoganisa* appear to be uncommon, only occasionally having been collected in larger numbers at light. Most specimens have been taken as singletons or in small numbers at any one time.

Preimaginal instars are still unknown. Both species have evidently never been reared, at least no descriptions of the larvae have been published.

Habitat preferences and ecology are also unknown. The imagines appear to mimic dead leaves and to inhabit primary and advanced secondary forest biotopes, which are especially endangered on the Philippines due to active and persisting deforestation.

Altitudinal data are few, as collecting records rarely include the elevation of the locality. The known altitudinal range for both species is from ca. 180 m to 2000 m, but it may extend further in both directions. This corresponds to mainly lowland and lower montane zones but also includes the lower regions of the upper montane zone. We have not detected obvious differences in altitudinal preferences between the two species so far.

Systematics

Genus Pseudoganisa Schultze, 1910

Schultze (1910: 162).

Type species: *Pseudoganisa currani* Schultze, 1910(: 162, text-fig. of venation; pl. 1, fig. 2), by original designation. — Genus originally described in family Eupterotidae; see Nässig & Oberprieler (2007, 2008).

After its original description, *Pseudoganisa* was evidently recognised as a separate genus in the printed literature only by Seitz (1922), Forbes (1955), Nässig (1995) and in the generic catalogue of Nässig & Oberprieler (2008), and then by Nässig & Treadaway (2009).

Tab. 2: Temporal patterns (i.e., specimens collected per month) of specimens of *Pseudoganisa currani* and *P. gonioptera* in CWAN, CCGT, SMFL, CMWM and CSLL (as far as a date is available for the specimens), separately for islands (from Nässig & Treadaway 2009, modified).

Species	Island	Jan.	Feb.	Mar.	Apr.	May	Jun.	Jul.	Aug.	Sep.	Oct.	Nov.	Dec.
Pseudoganisa currani	Mindanao	2	-	1	1	1	-	_	3	_	3	1	_
Pseudoganisa gonioptera	Luzon	_	3	3	1	_	2	1	_	1	2	-	-
	Mindoro	_	1	_	9	_	2	_	2	_	2	_	_
	Palawan	_	_	_	_	_	_	_	_	_	_	1	_
	Samar	2	_	_	_	_	_	_	_	_	1	_	_
	Leyte	_	2	1	_	4	1	1	1	5	_	_	_
Identity not clear (only few samples, no barcode available)	probably Pseudoganisa currani:												
	Dinagat	_	_	_	1	_	_	_	_	_	_	_	_
	Jolo	_	_	_	_	1	_	_	_	_	_	_	_
	probably Pseudoganisa gonioptera:												
	Panay	_	1	_	_	1	_	_	_	_	_	_	-
	Negros	_	8	1	5	7	2	2	4	1	1	2	2
	Cebu	_	_	-	_	_	1	2	_	-	_	_	_
	Bohol	_	1	-	_	_	_	_	_	_	_	_	_
	Total	4	16	6	17	14	8	6	10	7	9	4	2

Although its two now identified species, *P. currani* and *P. gonioptera*, are similar to the relatively homogeneous Asian species of *Apha* (and these two genera indeed appear to be closely related and most likely represent sistergroups), they are nevertheless sufficiently distinct in habitus and of genitalia (see Nässig & Treadaway 2009). In particular, the unusual structure of the of genitalia, especially:

- the apical part of the costal lobe of the valves is rolled around the needle-like apical part of the sacculus to form a sheath, and
- the gnathos arms are apparently fused to a weakly developed subscaphium in *Pseudoganisa*,

is unique among the Eupterotidae studied so far. The distribution of *Apha* is primarily continental, from the Himalaya to Indochina and China, but also includes some larger islands close to the Asian continent such as the Japanese archipelago as well as Taiwan and Hainan.

Pseudoganisa also superficially resembles species of Ganisa Walker, 1855 (as expressed by the generic name), but it is again very different in habitus and especially in ♂ genital structure (see Nässig & Treadaway 2009). Ganisa is much more widely distributed, from the Indian subcontinent across continental Indochina, southern and central China including Taiwan to the SE Asian islands of Sundaland, Sulawesi and Flores, also including the entire Philippine Archipelago; however, it, evidently, does not reach as far north as Apha.

The genus *Pseudoganisa* now contains the following two species:

Pseudoganisa currani Schultze, 1910

Schultze (1910: 162; text-fig. of venation; pl. 1, fig. 2).

Type material: ♂ holotype by original designation: "type no. 8748 in Entomological collection, Bureau of Science, Manila"; present depository unknown (possibly lost during the Japanese military occupation of Manila in world war II? At that time, and in the fighting and bombing during the return of US military forces, much scientific material was destroyed), not examined. — Type locality: Mindanao, District of Zamboanga, Port Banga, leg. W. I. HUTCHINSON. — Dedicated to a collector H. M. CURRAN, who "has contributed a large number of insects to our collection" (SCHULTZE 1910). Described in family Eupterotidae.

Distribution (see map, Fig. 10): Mindanao. — The populations from the islands of Dinagat (Mindanao subregion) and possibly also Jolo (Sulu region) have not been barcoded so far but possibly may also belong to *Pseudoganisa currani*.

No topotypical material of *P. currani* from the Zamboanga Peninsula (West Mindanao) was accessible to us, especially from the area of Zamboanga ("District of Zamboanga, Port Banga", the type locality) at the southern edge of this peninsula, due to acute problems of accessibility and civil disturbances.

Pseudoganisa gonioptera (West, 1932), stat. rev.

Apha gonioptera West (1932: 209-210; not illustrated).

Type material: ♂ holotype by original designation; in coll. A. E. WILEMAN in BMNH (examined by one of the authors [W.A.N.] and photographed by N. N. IGNATYEV; see NÄSSIG & TREADAWAY 2009: fig. 2). — Type locality: Luzon, subprov.

Benguet, Palali, 2000 ft., 2. III. 1913. — Named for its angulate wings. — West (1932: 210): "Nearest ally: *A. aequalis* Feld."; described in family Eupterotidae. — West evidently was unaware of Schultze's description and, consequently, did not compare with it.

= "Apha spec.?", undetermined: Semper (1896: 388; not illustrated), 1 \, damaged, "central Luzon", in SMFL (Nässig & Treadaway 2009: fig. 1); examined.

Distribution (see map): Probably all islands north of Mindanao; proven by barcode data for Luzon, Mindoro, Palawan, Leyte and Samar. — Material from all the other islands and regions has not yet been barcoded.

Note: This taxon *gonioptera* was evidently not dealt with in later printed literature except by Nässig (1995), Nässig & Oberprieler (2008) and Nässig & Treadaway (2009), but it is included in several name lists and biological name indexes in the web (Catalogue of Life 2012, Encyclopedia of Life 2012 etc., evidently all based on the Lepindex of BMNH 2005 [2013]), where it is always erroneously listed under *Apha* while its congener *currani* is correctly placed in *Pseudoganisa*. (For the general scientific value of such uncritical lists and indexes, which are not based on revisions of the taxa dealt with, see the comment in Nässig & Oberprieler 2008: 55.)

Pseudoganisa is readily distinguishable from Apha by its different wing shape, the latter genus generally having rounder wings except for a few species whose forewings are faintly "polygonal" (angulate or, more rarely, biangulate), and sexual dimorphism of wing shape is much weaker in Apha. Colour and pattern are somewhat similar (at least in some Apha species) but quite different in detail, as are the relations between wing shape and wing size compared to body size. In the ♂ genitalia the differences are even larger, as Apha has a bifid, basally fused uncus and its valves do not have a costal "sheath" around the sacculus. The gnathos of Apha is quite different from that of both Ganisa and Pseudoganisa, featuring only two well-developed, sclerotised lateral arms but no central plate, and the arms appear to be free from the rest of the body over their distal part.

Pseudoganisa differs from Ganisa by its wing shape (rounded in Ganisa, never angulate), ground colour (always light greyish-brown or dark greyish to blackish in Ganisa) and generally more intensive pattern, with fewer wavy lines in the median field than in Ganisa. In the & genitalia Ganisa also has a "bifid" uncus, with basally broadly fused lateral processes and often even a small rudiment

of the dorsal parts of the uncus detectable between them. It further has a well-developed gnathos, with the arms and plate fused and fully sclerotised and protruding caudad. In some species of the genus *Apona* (very closely related to *Ganisa*) the gnathos plate is medially elongated into a long, slightly bent, sword-like process extending even behind the valves.

Material of Pseudoganisa studied

For locality details, see the list of material examined in Nässig & Treadaway (2009) and additions below.

(We here only add specimens and locality data not included in the Nässig & Treadaway 2009 listing. The entire material is deposited especially in CCGT, CMWM, CSLL, CWAN and SMFL, with additions from a few other collections. For a complete list of all specimens studied by us until 2009, see the list of 2009. It appears that a specimen from "Sulawesi" in CMWM listed in 2009, now with GP Witt 11.581 = Ignatyev 006, is incorrectly labelled, as already stated in 2009; we do not expect that Sulawesi is also inhabited by *Pseudoganisa*.)

Largely from North to South, the Philippine Islands are grouped according to presently identified biogeographical regions (see Vane-Wright 1990, Treadaway 1995: maps pp. 12–13 and Treadaway & Schroeder 2012: map p. 6, fig. 3). The locality data contained in this list was the basis for plotting the distribution map, the collecting dates were included into Tab. 2.

1. Pseudoganisa gonioptera (West, 1932)

Luzon region:

Luzon (in total 15 $\eth \eth$, 1 \mathfrak{P}): 2 specimens barcoded.

Mindoro region:

Mindoro (in total 21 $\eth \eth$, 1 \mathfrak{P}): 1 specimen barcoded.

Palawan region:

Palawan (in total 1 \mathfrak{P}): 1 specimen barcoded.

West Visayan region:

Panay (in total 6 $\eth \eth$, 1 \lozenge).

Negros (in total $60 \, 33, 2 \, 99$).

Cebu (in total $3 \ \mathcal{F}(3)$).

Mindanao region:

East Visayan subregion:

Samar (in total 3 &&): 1 specimen barcoded.

Leyte (in total 41 $\eth \eth$, 1 \mathfrak{P}): 1 specimen barcoded.

Bohol (in total $1 \mathcal{Q}$).

2. Pseudoganisa currani Schultze, 1910

Mindanao region:

Mindanao subregion:

Mindanao (in total 34 &\$\mathrightarrow{\math

Dinagat (in total $1 \circ$).

Sulu region:

Sulu Archipelago, Jolo (in total 1 ♂).

Discussion

Even after the splitting into two species, *Pseudoganisa currani* as well as, especially, *P. gonioptera* are still externally quite variable.

The variability between islands (both in DNA and ♂ genitalia, as far as studied) of those populations of Ps. gonioptera which we have studied in more detail is comparatively large, with the populations from the eastern Visayan islands (Leyte and Samar) being rather distinct from the rest. However, the barcode of populations from so many islands has not yet been sequenced, and we expect that a more complete picture would show transitional steps in between. Also, the variabilty in of genitalia is also rather large and, especially, not fully coinciding with the presently known barcode variability. So we decided to keep all specimens from the islands north of Mindanao together in one species Pseudoganisa gonioptera, in spite of the observed, comparatively smaller differences. Further studies of genitalia morphology and DNA (perhaps also including other genes besides this COI fragment of the mitochondrial DNA) on more specimens from more islands should evaluate the status of the diverse islands populations in future.

Island records and zoogeography

Specimens of *Pseudoganisa* are known from all large and also from a few smaller islands of the Philippines (see map). The distribution area covers all biogeographical regions as defined by Vane-Wright (1990: 26), obviously including the Palawan area.

Most interestingly, *P. currani* is found only on Mindanao, i.e. in the Mindanao subregion of the Mindanao region *sensu* Vane-Wright (1990) (but possibly including Dinagat and Jolo? These populations have not yet been barcoded). It is, however, not found on the islands Samar and Leyte, being the East Visayan subregion of the Mindanao region *sensu* Vane-Wright.

With respect to *P. gonioptera*, the specimens from just these East Visayan islands (Leyte, Samar; not yet barcoded: Bohol) have a high barcode bootstrap value compared with the other populations. However, specimens from the West Visayan islands (and most other smaller islands in between and around) have not yet been barcoded, and the percentage of difference in sequence is (in spite of the high bootstrap value) much smaller towards the other northern islands than between the two species.

A specimen in CMWM shows a locality label "Indonesia, Sulawesi, Puncak Palopo". No specimen of the genus *Pseudoganisa* has ever been collected outside of the political unit of the Philippines. And further this part of Sulawesi is quite well-collected; any member of that genus should have been found by someone before. So we interprete this locality data as fully erroneous, being based on a wrong label attached to the specimen in error

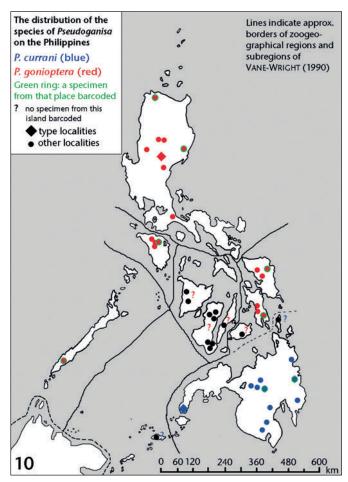


Fig. 10: Map of the Philippines with locality data of *Pseudoganisa* spp. (from Nässig & Treadaway 2009, modified and supplemented). (One dot may represent more than one locality in close proximity.) — Localities of the barcoded specimens marked with a green ring.

(compare Nässig & Treadaway 2009: 90, 91). Its genitalia do not offer a clear identification (Fig. 8), a barcode has not been achieved so far.

Hypothetic and speculative evolutionary scenario

This pattern may (on the very small basis barcoded so far!) perhaps tentatively suggest a colonisation of the Philippine Archipelago from the North, with the southern outliers having been subject to more "bottleneck events" than the northern ones during "island-hopping" during the spreading across the Philippine islands. A population of a member of an ancestor of today's genera *Apha* + *Pseudoganisa* might have been transported on one of the small microterranes today forming big parts of Palawan or Mindoro or other islands from close to the present-day Chinese coast of the Asian continent across the sea to the Philippines (see, e.g., HALL 1996, 1998 and other contributions in these books). This population developed its specific synapomorphies, especially in \mathcal{E} genitalia, during this long isolation. (A colonisation by active flight or winddrift is less likely, because the imagines evidently do not take up food and are rather short-lived.) This terrane-drifting took place during some time between mid-Oligocene (ca. 25 Ma b.p. [millions of years before present]) and early Pliocene (ca. 5 Ma b.p., when the microterranes agglomerated to the presentday Philippine Archipelago, according to HALL) — a long period surely sufficient for developing such strange synapomorphies. Later, when colonising the other islands of the Philippines and spreading there, the genetic diversification (expressed by the high variability of genitalia and also in the barcode) and later differentiation into distinct populations and eventually two species took place. However, more research is necessary on that subject.

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Entomologische Notiz

The name ‡Rueckbeilia is unpublished, unavailable and invalid for nomenclatural purposes presently, and it is an example of incorrect usage of the new Code Amendments especially by publisher and in online resources

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In 2012 (actually 10. ix. 2012, as stated in the journal website of the print journal "Cladistics"), an electronic prepublication in advance of the print version of a paper was made available to the readers. This prepublication (Talavera et al. 2012) is intended to be published in print within the printed version of "Cladistics" at some later time. It contains the description of a new name in the genusgroup of a blue butterfly (Lycaenidae), ‡Rueckbeilia.

Immediately after this, this name started to be used in several online databases (especially those containing DNA sequence data), with the authorship and publication date stated in the prepublication (for example: UniProt 2013, Comparative Toxicogenomic Database 2013, Immune Epitope Database and Analysis Resource 2013, and many others), and an empty page is already created even in the Wikspecies (2013) website. This name has also been cited and discussed with this publication date 2012 in many entomological internet forums.

Regrettably, the nomenclatural act of erecting this new genus-group name has not been validly published and made available under the ruling of the Code so far. The rules of Article 8.5.3 of the Code about electronic publications (ICZN 2012) have not been satisfied: the work containing the description of †*Rueckbeilia* was not registered in ZooBank before the prepublished electronic version was available An electronic prepublication from a print journal alone does not fulfill the requirements until it is physically published in print on paper. This means that the genus-group name †*Rueckbeilia* does not exist for the purpose of zoological nomenclature until now.

It is important here for nomenclature purposes to clearly distuinguish between entirely "electronic-only" publications (of which no printed version is generally existing, except someone prints the HTML or PDF file for his private use) and prepublications of print journals (which become available only when the print ver-